
Name of Organization: USGS-Biological Resources Division

Type of Organization: Federal Agency

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Project Title: Significance of two new exotic animals in Lake Michigan

Project Category: Exotic Species

Rank by Organization (if applicable): 0

Total Funding Requested (\$): 135,955 **Project Duration:** 2 Years

Abstract:

Two new exotic animals (Crustacea: Harpacticoida) have invaded the nearshore sands of southern Lake Michigan, and they are already the dominant harpacticoid species in these habitats. Preliminary sampling in limited areas of southern Lake Michigan indicated that exotics are *Schizopera borutzkyi*, a species previously known only from the Danube delta region of the Black Sea, and *Heteropsyllus* sp., likely a new species of this genus that was previously only represented in marine systems. Currently, nothing is known about the distribution of these exotics, or their seasonal fluctuations in abundance in Lake Michigan. Changes in the benthic crustacean community may cause drastic changes in the ecosystem, especially to higher trophic levels such as fish. We propose to use both 'extensive' (lake-wide) and 'intensive' (monthly) sampling of nearshore habitats to determine current distributions and seasonal fluctuations. In cooperation with other agencies, gut analyses will evaluate changes in feeding behavior (e.g., prey selection, capture success) of fish. Modern molecular techniques will be applied to establish the relationship among Lake Michigan species and closely related marine species, and potentially determine the arrival time of these exotics in Lake Michigan. This proposed work determines the history and extent of this new invasion and provides early indication of potential impacts of these invading species.

Geographic Areas Affected by the Project

States:

<input checked="" type="checkbox"/> Illinois	<input type="checkbox"/> New York
<input checked="" type="checkbox"/> Indiana	<input type="checkbox"/> Pennsylvania
<input checked="" type="checkbox"/> Michigan	<input checked="" type="checkbox"/> Wisconsin
<input type="checkbox"/> Minnesota	<input type="checkbox"/> Ohio

Lakes:

<input type="checkbox"/> Superior	<input type="checkbox"/> Erie
<input type="checkbox"/> Huron	<input type="checkbox"/> Ontario
<input checked="" type="checkbox"/> Michigan	<input type="checkbox"/> All Lakes

Geographic Initiatives:

<input checked="" type="checkbox"/> Greater Chicago	<input type="checkbox"/> NE Ohio	<input checked="" type="checkbox"/> NW Indiana	<input type="checkbox"/> SE Michigan	<input type="checkbox"/> Lake St. Clair
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Primary Affected Area of Concern: Not Applicable

Other Affected Areas of Concern:

For Habitat Projects Only:

Primary Affected Biodiversity Investment Area:

Other Affected Biodiversity Investment Areas:

Problem Statement:

Two new exotic species of benthic animal (Crustacea: Harpacticoida) have invaded the nearshore sands in southern Lake Michigan. One of the new invaders is *Schizopera borutzkyi* Montschenko, a species known only from the brackish waters of the Danube River delta area of the Black Sea (Montschenko 1967). The other invader likely is a new species belonging to the genus *Heteropsyllus*. Previously, this has been exclusively a marine and brackish water genus. The Lake Michigan species morphologically resembles *Heteropsyllus nunni* Coull, which is known from South Carolina and Nova Scotia coasts. B. Coull (University of South Carolina) and R. Huys (Natural History Museum of London) have independently suggested that this animal is a new species in this genus (personal communication). Modern molecular genetic techniques can be employed to determine relationships among the exotics in Lake Michigan and their congeners or sibling species in the native habitats, and potentially determine when the exotic arrived in Lake Michigan.

Based on current data, nothing is known about the distribution of these two exotics, their arrival time in Lake Michigan, or the impacts they may be exerting on the littoral or whole-lake ecosystem.

Objectives:

- 1) Establish the current distribution of both exotic species in Lake Michigan.
- 2) Describe the seasonal occurrence of the exotic crustaceans in order to understand their temporal influence on the ecosystem.
- 3) Determine the effects of the exotics on fish species.
- 4) Ascertain the genetic relationship between the *Heteropsyllus* sp. found in southern Lake Michigan and the closely related marine species (*Heteropsyllus nunni*).
- 5) Determine the feasibility of estimating the time of colonization of Lake Michigan.

Ecological impacts of these two species remain unknown. Preliminary evidence suggests that one or both of these new animals are the dominant harpacticoids in the areas studied. If further evidence is confirmed, then additional reductions of native species in other similar habitats in the Great Lakes could be expected. Ramifications of a shift in the species composition of benthic crustaceans could reverberate throughout the ecosystem. The ecological role of harpacticoids in the Great Lakes ecosystems is not well studied, but based on marine ecosystems, harpacticoids form an integral link in food webs by coupling nutrient inputs with higher trophic levels, such as salmonid fish (Fujiwara and Highsmith 1997), and also structure benthic and pelagic links (Walters and Shanks 1996). The freshwater studies outside of the Great Lakes suggest that harpacticoids are major consumers of daily carbon production (Perlmutter and Meyer 1991) and may provide an efficient pathway for energy transfer to higher trophic levels (Hudson et al. 1998). Whitman et al. (1994) demonstrated that harpacticoids numerically dominated (41%) the beach sands of southern Lake Michigan. Thus, dramatic changes in the harpacticoid community may cause drastic shifts in ecosystem processes originating in the benthic zone (Covich et al. 1999).

The greatest obstacle to determining the distribution and invasion history of these exotic species is that very few

studies have been conducted on the nearshore sand community in the Great Lakes, and especially in southern Lake Michigan (see Whitman et al. 1994). Nearly all studies in the nearshore habitats have focused on macroinvertebrates. Some early studies report harpacticoids in the genera *Canthocamptus* and *Bryocamptus* from nearshore sands in southeastern Lake Michigan (Evans and Stewart 1977, Nalepa and Quigley 1985). The most recent and intensive survey for harpacticoids relied mostly on fish-gut analyses, although some core samples were taken from nearshore sediments (Hudson et al. 1998). However, Hudson et al. (1998) rarely sampled sandy sediments and did not collect any samples from the southern basin of Lake Michigan. Based on the available literature, we have no historical data to pinpoint the initial invasion of either species. We have five years of data collected from sand communities in Indiana waters of Lake Michigan (Horvath et al. 2000, Last et al. 1995) and recently began expanding studies on the southwestern shore (Horvath et al. In prep.), but the distribution of both species remains unknown because of the limited scope and number of studies.

Proposed Work Outcome:

Lake-wide distribution (extensive sampling):

The lack of relevant data on the harpacticoids in Lake Michigan, coupled with the small amount of preliminary data available, makes it currently impossible to determine the extent of the invasion by the two new exotics. We, therefore, will implement an extensive survey of nearshore benthic habitats to map the current distribution of these exotic crustaceans. Extensive sampling sites will be chosen based on the known distribution of the exotics. Sampling intensity will decrease exponentially away from the known distribution. This will maximize our ability to determine the rate of dispersal of the exotics as well as contribute to the distribution maps. Extensive sites will be visited in July and November to represent summer and winter distributions. Sands will be sampled by ponar or SCUBA. Sands will be elutriated per Whitman et al. (1983). Harpacticoids will be identified and voucher specimen formally archived.

Seasonal population dynamics (intensive sampling):

Heteropsyllus spp. from marine habitats are considered univoltine winter species that encyst during summer (Williams-Howze and Coull 1992). However, our preliminary samples indicate that adult *Heteropsyllus* sp. is found in appreciable numbers from July to at least December. There is no report of *S. Borutzkyi* seasonal dynamics in the literature. These data indicate that marine life histories are not easily extrapolated to Lake Michigan. It is, then, necessary to establish when these exotic species are most abundant in Lake Michigan, which potentially will indicate when they exert their greatest impact on the benthic and benthic-linked ecosystems. Continuous year-round sampling will provide the adequate data to determine seasonal abundances.

Based on the initial results from the extensive sampling, at least 2 sites will be chosen to be intensive sampling sites. Choice of sites will consider the existent populations and accessibility. Intensive sites will be sampled during ice-free months. Evidence of encystment will be sought for comparison to marine congeners.

Effects on higher trophic levels:

Shifts in the species composition of crustaceans could have effects on higher trophic levels, especially if behavior is greatly different between the new and native species. Fish species that rely on native harpacticoids may not be able to capture the exotic species if they exhibit greater cryptic behaviors (Gee 1989; Shaw and Jenkins 1992). Such effects can be evident in changes in fish feeding, which can be quantified by changes in gut content to prey availability. For example, if sculpin guts contain mostly native harpacticoid species when natives comprise only a small percentage of the harpacticoids in the field, then we may conclude that the dominant exotics are not being captured as efficiently. Conversely, if the exotics are found in similar proportions in guts and in the field, then we may conclude that the fish can capture these animals with equal efficiencies, and thus would not expect large ecological consequences.

In order to determine the possible effects of a shift in the species composition of benthic crustaceans on higher trophic levels, we will examine fish gut content from fish collected in areas known to be inhabited by the two exotic animals. Bottom-feeding fish species or species that occasionally bottom feed include sculpins, sticklebacks, alewife, chub, and smelt. These species consume benthic copepods in large numbers (Hudson et al. 1995) and the copepods remain identifiable (Hudson et al. 1998).

In collaboration with state Department of Natural Resources (DNRs), fish will be collected and gut content examined. Fish will be collected in routine trawl samples conducted by the DNRs. Appropriate fish species will be preserved in formalin until guts can be removed and their contents examined. Comparisons to background prey densities will be made to determine preferential feeding.

Genetic analyses:

We plan to look at patterns of genetic variation among individuals sampled from each species to establish the relationships

among the species of interest. Analyses of variation in genetic material provides an objective method for reconstructing evolutionary history based on shared attributes among closely related species. We may also be able to determine how long the new species have been in Lake Michigan. This is feasible if there is an underlying assumption that the unknown species and *S. borutzkyi* are recent colonists or if it is known that the marine and freshwater species have been segregated.

Total genomic DNA will be extracted from preserved specimens of *H. nunti* and the Lake Michigan *Heteropsyllus* sp. using the protocol of Schizas et al. 1997. Individual copepods will be incubated in 10 µl of cold PCR Buffer II with 1 µl of 1 mg/ml Proteinase K at 55°C for 3 hours. After heat inactivation of the Proteinase K, 10 µl of GeneReleaser will be added to the mixture. After centrifugation, the supernatant containing the total genomic DNA will be used directly as the substrate for in vitro amplification of DNA with the polymerase chain reaction (PCR). We plan to use a combination of PCR and restriction fragment length polymorphism analysis (PCR-RFLP) to survey genetic variation in the mitochondrial DNA genome. Mitochondrial DNA (mtDNA) is a rapidly evolving molecule that is inherited monoclally, and is transmitted through the maternal lineage in a population. Advantages to using this molecule are that mtDNA is present in a high copy number per cell, which provides a large number of starting copies of DNA, and it is transmitted to offspring intact and is not subject to the effects of recombination.

The gene loci that we plan to survey include cytochrome c oxidase I and cytochrome b, both of which have been used successfully for population genetic analyses of metazoan invertebrates (e.g. Folmer et al. 1994). We will attempt to survey additional variable regions as needed (e.g., the nuclear gene coding for histone I per Burton (1998) or the control region (D-loop) for sequence variation). The control region of mtDNA is a highly variable non-coding region in most species, and is the region at which replication of the mtDNA molecule occurs. Primer design will be greatly facilitated by similar molecular work on marine copepods.

Literature cited:

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Project Milestones:

Dates:

Project Start	06/2000
Field sampling, molecular work	07/2000
End field sampling, manuscript prep.	01/2001
Resume field sampling	03/2001
End field sampling, manuscript prep.	01/2002
Resume field sampling	03/2002
Manuscript prep.	04/2002
Project End	05/2002

☐ Project Addresses Environmental Justice

If So, Description of How:

☐ Project Addresses Education/Outreach

If So, Description of How:

Project Budget:

	Federal Share Requested (\$)	Applicant's Share (\$)
Personnel:	68,172	50,000
Fringe:	17,043	17,000
Travel:	10,000	3,000
Equipment:	3,000	2,000
Supplies:	3,000	1,000
Contracts:	5,000	0
Construction:	0	0
Other:	0	0
Total Direct Costs:	106,215	73,000
Indirect Costs:	29,740	0
Total:	135,955	73,000
Projected Income:	0	0

Funding by Other Organizations (Names, Amounts, Description of Commitments):

Army Corps of Engineers - \$25k - Collection of sand samples for meiobenthos composition.

Description of Collaboration/Community Based Support:

Fish sampling will be conducted/coordinated with the cooperation of Departments of Natural Resources from the states of Indiana, Illinois, Michigan, and Wisconsin. They also will provide access to Lake Michigan at our intended sampling sites.

Sleeping Bear National Lakeshore will provide sampling assistance in northern Lake Michigan.

Indiana Dunes National Lakeshore will provide sampling assistance in southern Lake Michigan.

Ship time on the R/V Lake Guardian will be requested.

Dr. Mary Burnham Curtis at the Great Lakes Science Center is a co-PI and will be overseeing the molecular laboratory work and data analyses.

Dr. Thomas Horvath at Lake Michigan Ecological Research is a co-PI and will be involved in field work, data analyses, and manuscript preparation.